Amendments to the Specification:

Please add the following new paragraphs after the paragraph ending on page 6, line 21:

The Hutchinson strain (Hutch) of non-A, non-B hepatitis virus (NANBV) has been propagated through passage in animals and portions of the virus have been cloned and sequenced. Sequence data shows differences at both the nucleotide and amino acid level when compared to any previously reported NANBV strains. See, for comparison, Okamoto, et al., Japan J. Exp. Med., 60:163-177 (1990); and International Application No. PCT/US88/04125.

The identified sequences have been shown herein to encode structural proteins of NANBV. The NANBV structural proteins are also shown herein to include antigenic epitopes useful for diagnosis of antibodies immunoreactive with structural proteins of NANBV, and for use in vaccines to include neutralizing antibodies against NANBV.

The nucleotide sequence that codes for the amino terminal polyprotein portion of the structural genes of the Hutch strain of NANBV is contained in Figure 9. By comparison to putative relatives of NANBV, namely to other NANBV isolates, to flavivirus, and to pestivirus, the nucleotide sequence contained in Figure 9 is believed to encode structural proteins of NANBV, namely capsid and portions of envelope.

The structural antigens described herein are present in the putative capsid protein contained in Figure 9 from amino acid residue positions 1-120, and are present in the amino terminal portion of the putative envelope protein contained in Figure 9 from residue positions 121-326.

The present invention contemplates a DNA segment encoding a NANBV structural protein that comprises a NANBV structural antigen, preferably capsid antigen. A particularly preferred capsid antigen includes an amino acid residue sequence represented by Figure 9 from residue 1 to residue 20, from residue 21 to residue 40, from residue 2 to residue 40, or from residue 1 to residue 74, and the DNA segment preferably includes the nucleotide base sequence represented by Figure 9 from base position 1 to base position 60, from base position 61 to base position 120, from base position 4 to base position 120, or from base position 1 to base position 222, respectively.

Also contemplated is a recombinant DNA molecule comprising a vector, preferably an expression vector, operatively linked to a DNA segment of the present invention. A preferred recombinant DNA molecule is pGEX-3X-690:691, pGEX-3X-690:694, pGEX-3X-693:691, pGEX-3X-15:17, pGEX-3X-15:18, pGEX-2T-15:17, pGEX-2T-CAP-A, pGEX-2T-CAP-B or pGEX-2T-CAP-A-B.

A NANBV structural protein is contemplated that comprises an amino acid residue sequence that defines a NANBV structural antigen, preferably a capsid antigen, and more preferably one that includes the amino acid residue sequence contained in Figure 9 from residue 1 to residue 20, from residue 21 to residue 40, from residue 2 to residue 40, or from residue 1 to residue 74. Fusion proteins comprised of a NANBV structural protein of this invention are also contemplated.

Further contemplated is a culture of cells transformed with a recombinant DNA molecule of this invention and methods of producing a NANBV structural protein of this invention using the culture.

Also contemplated is a composition comprising NANBV structural protein. The composition is preferably characterized as being essentially free of (a) procaryotic antigens, and (b) other NANBV-related proteins.

Still further contemplated is a diagnostic system in kit form comprising, in an amount sufficient to perform at least one assay, a NANBV structural protein composition of this invention, as a separately packaged reagent.

In another embodiment, the present invention contemplates a diagnostic system, in kit form, comprising a fusion protein of this invention. Preferably, the diagnostic systems contains the fusion protein affixed to a solid matrix.

Further contemplated is a method of assaying a body fluid sample for the presence of antibodies against at least one of the NANBV structural antigens described herein. The method comprises forming an immunoreaction admixture by admixing (contacting) the body fluid sample with a fusion protein of this invention. The immunoreaction admixture is maintained for a time period sufficient for any of the antibodies present to immunoreact with the fusion protein to form an immunoreaction product, which product, when detected, is indicative of the presence of anti-NANBV structural protein antibodies. Preferably, the fusion protein is affixed to a solid matrix when practicing the method.

In another embodiment, this invention contemplates a vaccine comprising an immunologically effective amount of a NANBV structural protein of this invention in a pharmaceutically acceptable carrier. The vaccine is essentially free of (a) procaryotic antigens, and (b) other NANBV-related proteins.

A prophylactic method for treating infection, which method comprises administering a vaccine of the present invention, is also contemplated.

Please add the following new paragraphs after the paragraph ending on page 43, line 15, and after the Supplemental Amendment to the Specification, filed February 13, 2006, page 5, line 1 to page 51, line 14:

pGEX-2T-CAP-A: Oligonucleotides 1-20 (+) and 1-20 (-) for constructing the vector pGEX-2T-CAP-A for expressing the CAP-A fusion protein were prepared as described in Example 9A(2) having nucleotide base sequences corresponding to Figure 14 and Figure 15, respectively.

Oligonucleotides 1-20 (+) and 1-20 (-) were admixed in equal amounts with the expression vector pGEX-2T (Pharmacia) that had been predigested with Eco RI and Bam HI and maintained under annealing conditions to allow hybridization of the complementary oligonucleotides and to allow the cohesive termini of the resulting double-stranded (ds) oligonucleotide product to hybridize with pGEX-2T at the Eco RI and Bam HI cohesive termini. After ligation the resulting plasmid designated pGEX-2T-CAP-A contains a single copy of the ds oligonucleotide product and a structural gene coding for a fusion protein designated CAP-A having an amino acid residue sequence shown in Figure 11 from residue 1 to residue 252.

The pGEX-2T vector is similar to the pGEX-3X vector described above, except that the resulting fusion protein is cleavable by digestion with the site specific protease thrombin.

pGEX-2T-CAP-B: Oligonucleotides 21-40 (+) and 21-40 (-) for constructing the vector pGEX-2T-CAP-B for expressing the CAP-B fusion protein were prepared as described in Example 9A(2) having nucleotide base sequences corresponding to Figure

16 and Figure 17, respectively.

Oligonucleotides 21-40 (+) and 21-40 (-) were admixed in equal amounts with the pGEX-2T expression vector that had been predigested with Eco RI and Bam HI and maintained under annealing conditions to allow hybridization of the complementary oligonucleotides and to allow the cohesive termini of the resulting double-stranded oligonucleotide product to hybridize with pGEX-2T at the Eco RI and Bam HI cohesive termini. After ligation the resulting plasmid designated as pGEX-2T-CAP-B contains a single copy of the ds oligonucleotide product and contains a structural gene coding for a fusion protein designated CAP-B having an amino acid residue sequence shown in Figure 12 from residue 1 to residue 252.

pGEX-2T-CAP-A-B: Oligonucleotides for constructing the vector pGEX-2T-CAP-A-B for expressing the CAP-A-B fusion protein were prepared as described in Example 9A(2) having nucleotide base sequences corresponding to Figure 18 and Figure 19, respectively.

Oligonucleotides according to Figure 18 and Figure 19 were admixed in equimolar amounts with the plasmid pGEX-3X-690:694 described in Example 9B(2). The admixture was combined with the reagents for a polymerase chain reaction (PCR) and the two admixed oligonucleotides were used as primers on the admixed pGEX-3X-690:694 as template in a PCR reaction to form a PCR extension product consisting of a

double-stranded nucleic acid molecule that encodes the amino acid residue sequence contained in Figure 9 from residue 2 to 40 and also includes PCR-added restriction sites for Bam HI at the 5' terminus and Eco RI at the 3' terminus. The PCR extension product was then cleaved with the restriction enzymes Bam HI and Eco RI to produce cohesive termini on the PCR extension product. The resulting product with cohesive termini was admixed in equal amounts with the pGEX-2T expression vector that had been predigested with Eco RI and Bam HI and maintained under annealing conditions to allow the cohesive termini of the double-stranded PCR extension product to hybridize with pGEX-2T at the Eco RI and Bam HI cohesive termini. After ligation the resulting plasmid designated pGEX-2T-CAP-A-B contains a single copy of the double-stranded PCR extension product and contains a structural gene coding for a fusion protein designated CAP-A-B having an amino acid residue sequence shown in Figure 13 from residue 1 to residue 271.

In Table 7, comparative results are presented from anti-HCV capsid fusion protein assays according to the basic immunoblot assay described in Example 12 using various chimp and human sera on the following HCV capsid fusion proteins: CAP-N, CAP-A, CAP-B and CAP-C.

TABLE 7

SERA	TYPE[a]	CAP-N[b]	CAP-A[c]	CAP-B[d]	CAP-C[e]
C18	Chimp 10 (A)	+++	+	+	-
C10	Chimp 194 (A)	+++	+++	+++	-
59-16	Chimp 59 (A)	+++	+	+++	ND
59-12	Chimp 59 (A)	ND[f]	++	+++	-
C9	Chimp 181 (A)	+++	-	+++	-
213-18	Chimp 213 (A)	ND	+	+	-
C2	Chimp 10 (C)	++	-	-	-
C1	Chimp 10 (C)	+++	-	-	-
C19	Chimp 10 (C)	+++	-	-	-
C4	Chimp 68 (C)	+++	+++	+++	ND
169-16	Human	ND	+++	+++	-
169-23	Human	ND	+++	+++	-
191-1	Human	+	+	+	ND
191-2	Human	+	+	++	ND
191-3	Human	+	+	+	ND
216-1	Human	-	+/-	+/-	ND
216-2	Human	+	+	+	ND
216-3	Human	+	+	+	ND

a The type of sera tested is indicated by the species (chimp or human), a chimp identification number if the sample is from a chimp, and a designation (in parenthesis) if the sera donor exhibits acute (A) or chronic (C) HCV infection at the time the sera was sampled.

b CAP-N indicates the GST:NANBV 690:694 fusion protein produced in Example 13 that includes HCV capsid protein residues 1-74.

c CAP-A indicates the GST:NANBV fusion protein produced in Example 13 that includes HCV capsid protein residues 1-20.

d CAP-B indicates the GST:NANBV fusion protein produced in Example 13 that includes HCV capsid protein residues 21-40.

e CAP-C indicates the GST:NANBV fusion protein produced in Example 13 that includes HCV capsid protein residues 41-60.

f +, ++ and +++ indicate relative amounts of anti-HCV capsid antibody immunization product detected by the western blot assay, where + indicates a weak band after overnight exposure of the x-ray film, ++ indicates a strong band after overnight exposure of the x-ray film, +++ indicates a strong band after 1 to 2 hours exposure of the x-ray film, and +/- or - indicates a faint or no band, respectively, after overnight exposure of the x-ray film.

g "ND" indicates not tested.

The results shown in Table 7 indicate that fusion proteins containing the CAP-A antigen or CAP-B antigen are immunoreactive with antibodies present in sera from HCV-infected humans or chimps. In addition, CAP-C antigen does not significantly immunoreact with sera from HCV infected humans or chimps.

Other GST:NANBV fusion proteins described herein were also expressed in cultures of E. coli Strain W3110 as described above using the GST fusion protein vectors produced in Example 9 after their introduction by transformation into the E. coli host. After induction and lysis of the cultures, the GST fusion proteins were purified as described above using glutathione agarose affinity chromatography to yield greater than 95% pure fusion protein as determined by SDS-PAGE. Thus, CAP-A, CAP-B and CAP-C fusion proteins were all expressed and purified as above using the pGEX-2T-CAP-A vector, the pGEX-2T-CAP-B vector, or the pGEX-2T-CAP-C vector, respectively, and CAP-A-B fusion protein is expressed and purified using the PGEX-2T-CAP-A-B vector.